No Deterioration in Insulin Sensitivity, but Impairment of both Pancreatic β-Cell Function and Glucose Sensitivity, in Japanese Women with Former Gestational Diabetes Mellitus

H. Sakamaki[1], H. Yamasaki[1], K. Matsumoto[2], K. Izumino[1], H. Kondo[1], Y. Sera[1], M. Ozaki[1], T. Abe[1], E. Kawasaki[2], H. Takino[1], Y. Yamaguchi*[1], K. Eguchi[1]

[1]*The First Department of Internal Medicine, Nagasaki University School of Medicine, Nagasaki 852–8501, Japan*
[2]*Sasebo-chuou Hospital, Sasebo-city, Nagasaki 857–11, Japan*

To identify the primary pathogenic factors involved in the development of Type 2 diabetes mellitus (DM), we studied Japanese women with former gestational diabetes mellitus (GDM) who are at risk for the later development of Type 2 DM. We used the minimal model analysis derived from frequently sampled intravenous glucose tolerance test (FSIGT). The subjects consisted of eight non-obese women with a history of GDM and eight non-obese normal women as control subjects. The 75 g oral glucose tolerance test (75 g OGTT) performed within 6 months of delivery confirmed that all the subjects with former GDM had a normal glucose tolerance. Insulin sensitivity (SI) derived from the minimal model analysis was not different between the two groups. Glucose effectiveness at zero insulin (GEZI), reflecting tissue glucose sensitivity, was significantly lower in former GDM patients than in control subjects (1.18 ± 0.34 vs $2.26 \pm 0.29 \times 10^{-2}\,\mathrm{min}^{-1}$, $p < 0.05$). The early phase insulin secretion found in FSIGT was markedly reduced to 56 % of that observed in control subjects (1250 ± 187.4 vs $2223 \pm 304.3\,\mathrm{pmol\,l^{-1}\,min}$, $p < 0.01$). Our results indicate that in former GDM patients, who are Japanese and non-obese, impairment of the acute insulin response to glucose and a decrease in tissue glucose sensitivity rather than insulin sensitivity are the primary pathogenic factors involved. © 1998 John Wiley & Sons, Ltd.

Diabet. Med. **15: 1039–1044 (1998)**

KEY WORDS **gestational diabetes mellitus; insulin sensitivity; β-cell function**

Received 24 March 1998; revised 17 July 1998; accepted 17 July 1998

Introduction

Type 2 diabetes mellitus (DM) is pathophysiologically characterized by deterioration of β-cell function and/or tissue sensitivity to insulin.[1–5] Women with a recent history of gestational diabetes mellitus (former GDM) are reportedly at high risk for future development of Type 2 DM, with a 15–47 % increased risk of developing the disease within 5 years after child birth.[6,7] Results of several studies examining Caucasian women with former GDM suggest that impairment of a compensatory secretion of insulin against reduced insulin sensitivity is the pathogenic mechanism.[8–12]

However, impairment of β-cell insulin secretion is one of the important characteristics of Japanese patients with impaired glucose tolerance (IGT) or Type 2 DM[13–15] and Japanese offspring of patients with Type 2 DM.[16] However, to our knowledge, metabolic parameters including β-cell response to glucose, tissue insulin sensitivity, and glucose sensitivity, have not been previously examined in former GDM Japanese female patients with normal glucose tolerance.

In the present study, we examined non-obese Japanese women who had normal glucose tolerance but a history of GDM and evaluated glucose metabolism, using the minimal model analysis from an insulin-modified FSIGT to identify the primary pathogenic factor(s) that may lead to the development of Type 2 DM.

Subjects and Methods

Eight non-obese (body mass index $<26.0\,\mathrm{kg\,m^{-2}}$) women with former GDM participated in the present study. During pregnancy, all subjects were tested with 75 g

Abbreviations: AIR acute insulin response, BIE basal insulin effect, GEZI glucose effectiveness at zero insulin, I_o basal insulin, FSIGT frequently samped ultravenous glucose tolerance, SG glucose sensitivity, SI insulin sensitivity

* Correspondence to: Dr Yoshihiko Yamaguchi, The First Department of Internal Medicine, Nagasaki University School of Medicine, 1–7–1 Sakamoto, Nagasaki 852–8501, Japan E-mail:yama@net.nagasaki-u.ac.jp

CCC 0742–3071/98/121039–06$17.50
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OGTT and were diagnosed as IGT according to WHO criteria.[6] They were subjected to 75 g OGTT again and insulin modified FSIGT to estimate insulin sensitivity and β-cell function no less than 6 months after each delivery. We also studied eight non-obese normal subjects as control subjects who had no endocrine disease, liver disease, hypertension, family history of diabetes mellitus (Type 1 and Type 2), had had normal glucose levels both during pregnancy and after delivery confirmed by 75 g OGTT, and were not on any medications. A written informed consent was obtained from each subject and the study protocol was approved by the Ethics Review Committee of Nagasaki University School of Medicine.

Insulin-modified FSIGT

FSIGT was performed while the patient was supine after 12 h of fasting, during the follicular phase of the menstrual cycle. Baseline samples were obtained at −20, −10 and −3 min before glucose (50 % dextrose) was administered intravenously at a dose of 300 mg kg^{-1} body weight over 1 min and 27 subsequent blood samples were obtained. Soluble insulin (Humalin R; Shionogi, Osaka, Japan) at a dose of 20 mU kg^{-1} body weight was infused into another antecubital vein, 20 to 25 min after the administration of glucose as described previously by our group.[17,18]

Analytical Methods

Plasma glucose was measured in duplicate with an automatic analyser (Kyoto-Daiichi-Kagaku, Kyoto, Japan) by a glucose oxidase method. Immunoreactive insulin (IRI) was measured in duplicate using a Phadeseph insulin RIA kit (Shionogi, Osaka, Japan).

Data Analysis

Glucose disappearance rate (Kg) was calculated as the slope of the least-square regression line relating the natural logarithm of glucose concentration to time using five samples withdrawn between 10 and 19 min. Insulin sensitivity (SI) and glucose effectiveness (SG) were estimated by the minimal model approach.[19,20] The basal insulin effect (BIE) is the product of basal insulin (I_0b) and SI: BIE = $I_o \times$ SI. Glucose effectiveness at zero insulin (GEZI) is the difference between SG and BIE, i.e. GEZI = SG − ($I_o \times$ SI). This measure is analogous to tissue glucose sensitivity.[21,22]

The acute endogenous insulin response (AIR) to glucose was expressed as the integrated area under the insulin curve above the basal level between 0 and 20 min in FSIGT. In this study, we calculated the disposition index (DI) as a product of SI × AIR, which is constant in healthy individuals.[23,24]

Statistical Analysis

Statistical analyses were performed by two-tailed Student's *t*-test. Data were expressed as mean ± SEM. A p value <0.05 was considered significant.

Results

Clinical Characteristics

The clinical characteristics of our subjects are shown in Table 1. The mean age, height, weight, body mass index (BMI) and waist to hip ratio of the former GDM subjects were similar to those of the control group. None of the subjects with former GDM had antibodies to glutamic acid decarboxylase (GAD) antibody or islet cell cytoplasm (ICA).

75 g OGTT

The mean values of plasma glucose concentrations and IRI levels during the 75 g OGTT performed during the first 6 months after delivery are shown in Figure 1. Plasma glucose concentrations in former GDM indicated normal glucose tolerance according to WHO criteria but glucose levels at 90 and 120 min were significantly higher than those of controls. The IRI at 120 min was significantly higher in former GDM and the peak time was delayed compared to those of control subjects. In former GDM, the area under the curve above basal glucose ($AUC_{glucose}$) was not significantly different from control (375.7 ± 82.6 vs 207.2 ± 62.3 mmol l^{-1} min, p = 0.15) and that of IRI (AUC_{IRI}) was significantly higher than control subjects (39 359 ± 7220 vs 14 143 ± 3730 pmol l^{-1}min, $p < 0.05$).

FSIGT and Minimal Model Analysis

Mean plasma glucose and insulin concentrations during FSIGT are shown in Figure 2 and the results of minimal model analysis from FSIGT in Table 2. Kg was significantly lower in former GDM than control subjects (1.89 ± 0.34 vs 2.94 ± 0.27 % min^{-1}, $p < 0.05$). SI was not significantly different in former GDM from that in control subjects. However, AIR to glucose in former GDM was

Table 1. Clinical characteristics of former GDM and control subjects

	Control subjects ($n = 8$)	Former GDM ($n = 8$)
Age (yr)	35.4 ± 2.4	40.5 ± 2.0
Height (cm)	155.3 ± 1.6	152.8 ± 2.2
Weight (kg)	51.6 ± 0.8	51.1 ± 2.5
BMI (kg m^{-2})	21.4 ± 0.4	21.9 ± 1.0
Waist to hip ratio	0.72 ± 0.02	0.79 ± 0.03

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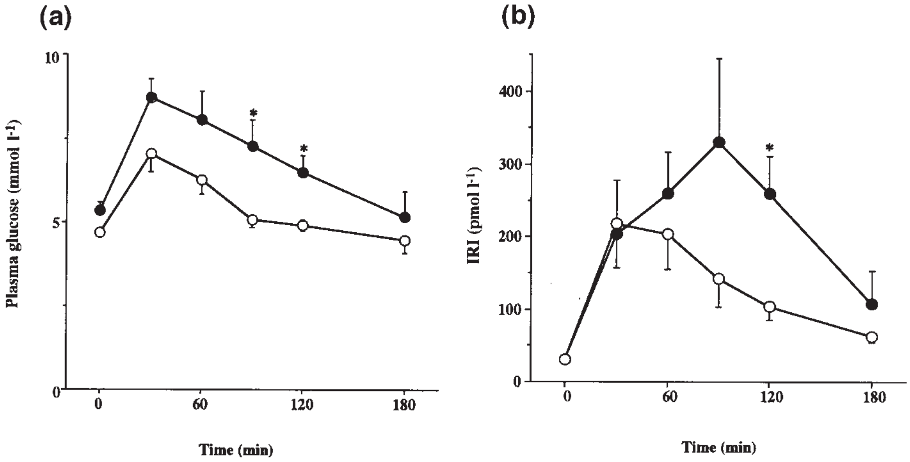

Figure 1. (a) Plasma glucose and (b) insulin concentrations measured during 75 g OGTT in women with former GDM (closed circles) and control subjects (open circles); *$p < 0.05$ vs control subjects

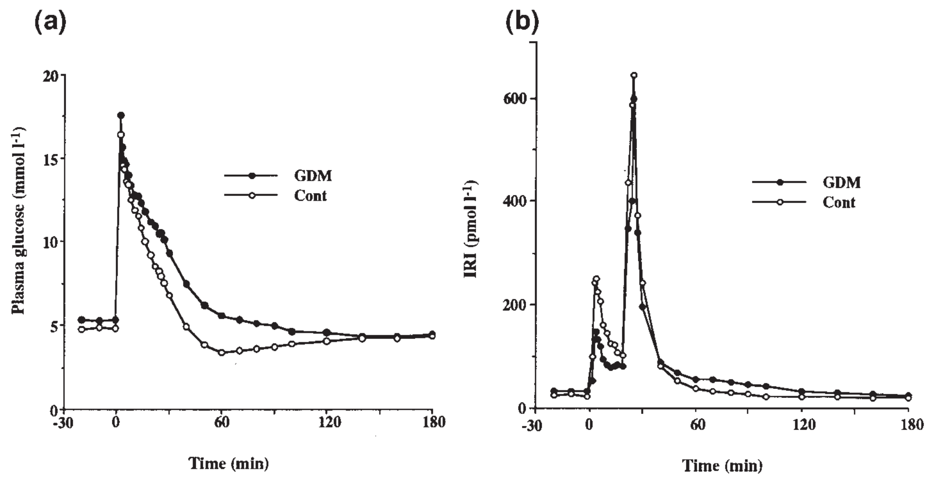

Figure 2. Serial changes in (a) mean plasma glucose and (b) insulin concentrations during modified FSIGT in women with former GDM (closed circles) and control subjects (open circles). The reduced rate of change in plasma glucose toward the basal level in former GDM was slower than that in control subjects. The acute insulin response (AIR) to glucose (insulin area during 0–20 min) in former GDM was reduced by 44 % compared with that in control subjects

Table 2. Results of minimal model analysis from FSIGT

	Control	Former GDM	p value
Basal glucose (Gb) (mmol l^{-1})	4.78 ± 0.14	5.24 ± 0.39	0.29
Basal insulin (I_o) (pmol l^{-1})	24.6 ± 3.4	30.9 ± 4.4	0.27
Kg (% min^{-1})	2.94 ± 0.27	1.89 ± 0.34	0.028[a]
SI ($\times 10^{-4}$ $pmol^{-1}$ min^{-1} l)	2.70 ± 0.32	3.26 ± 0.77	0.52
SG ($\times 10^{-2}$ min^{-1})	2.97 ± 0.41	2.13 ± 0.49	0.19
GEZI ($\times 10^{-2}$ min^{-1})	2.26 ± 0.29	1.18 ± 0.34	0.029[a]
BIE ($\times 10^{-2}$ min^{-1})	0.71 ± 0.18	0.95 ± 0.25	0.42
AIR (pmol l^{-1} min)	2223.4 ± 304.3	1250.2 ± 187.4	0.0094[b]
DI ($\times 10^{-4}$)	5023.5 ± 313.3	3474.2 ± 578.7	0.029[a]

[a] $p < 0.05$ vs control subjects.
[b] $p < 0.01$ vs control subjects.

significantly lower than in control subjects (1250 ± 187 vs 2223 ± 304 pmol l^{-1} min, $p < 0.01$). SG in former GDM tended to be lower compared with control subjects but the difference was not significant. GEZI in former GDM was significantly lower than in control subjects ($1.18 \pm 0.34 \times 10^{-2}$ vs $2.26 \pm 0.29 \times 10^{-2}$ min^{-1}, $p < 0.05$), although there was no difference in BIE between the two groups (Table 1, Figure 3). The

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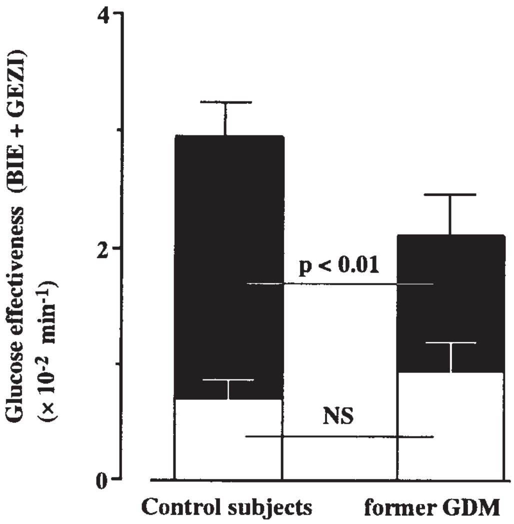

Figure 3. The values of SG were divided into GEZI component (solid bars) and BIE component (open bars). In former GDM, GEZI was significantly lower than in control subjects, while SG (GEZI + BIE) and BIE were not significantly different from control subjects

disposition index (SI × AIR) in former GDM was lower than in control subjects ($3474 \pm 578 \times 10^{-4}$ vs $5024 \pm 313 \times 10^{-4}$, $p < 0.05$) (Table 1). Relationships between SI and AIR to glucose is illustrated in Figure 4. The curve constructed by each set of data showed a downward shift in former GDM as reflected by a decrease in AIR.

Discussion

Our results showed that in non-obese Japanese women with former GDM, insulin sensitivity (SI) was not different from control subjects although the insulin response to a glucose challenge was markedly attenuated. Doi *et al.*[16] and Taniguchi *et al.*[25] demonstrated that non-obese Japanese subjects with IGT and the offspring of Type 2 DM parents with normal glucose tolerance have reduced early phase insulin secretion but normal insulin sensitivity. Taken together, our results indicate that impairment of pancreatic β-cell function in former GDM patients may be an important pathogenic factor in development of Type 2 DM in female Japanese. In our subjects, the insulinogenic index, which was defined as the ratio of the increment of insulin to that of glucose in 75 g OGTT ($\Delta IRI_{0\text{–}30\ min}/\Delta$plasma glucose $_{0\text{–}30\ min}$), was not diminished (data not shown), and the insulin area under the curve during 75 g OGTT was even significantly increased. The reason for the difference in β-cell response between FSIGT and 75 g OGTT in our study is that a normal insulin response during oral glucose tolerance test does not necessarily exclude the impairment of β-cell function, as demonstrated by Pimenta *et al.*[2] Alternatively, the difference may be due to the small population sample in our study. In the present study, the disposition index was also significantly reduced in subjects with former GDM, suggesting impairment of acute β-cell response to glucose.

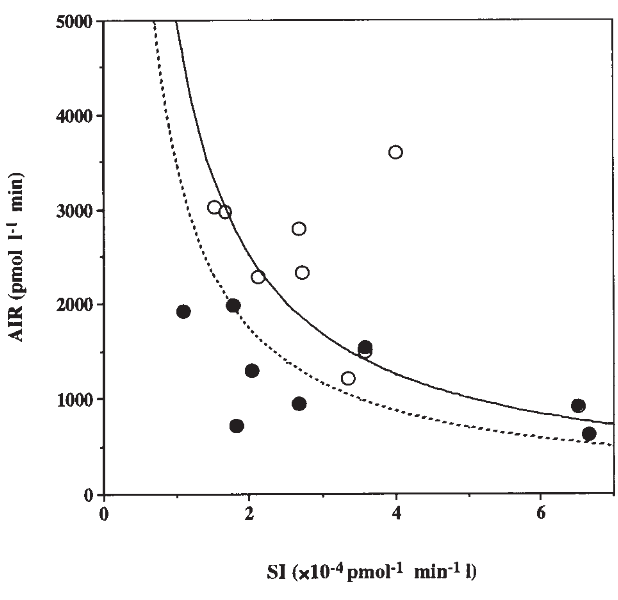

Figure 4. Relationship between SI and AIR to glucose. The solid and broken lines are drawn for control subjects and former GDM, respectively. The regression curve of former GDM was shifted below that of control subjects

In our subjects with former GDM, glucose effectiveness at zero insulin (GEZI) was significantly reduced compared with control subjects. Similar results were reported by Taniguchi *et al.*,[25,26] demonstrating that Japanese patients with IGT had impaired β-cell function as well as reduced glucose sensitivity (SG) and (GEZI). On the other hand, Finegood and Tzur[27] indicated that insulin secretory function itself influenced estimates of SG. However, we have found a lack of significant correlation between the acute insulin response and SG in the present as well as previous studies.[17,18] Based on these findings, we postulate that Japanese subjects probably suffer from impairment of β-cell function and glucose-dependent glucose uptake.

In contrast, in Caucasian and Pima Indian subjects, several investigators have suggested that insulin resistance is the primary defect in Type 2 DM, while impairment of insulin secretion is a secondary defect.[3–5] Using the minimal model analysis, Byrne *et al.*[9] demonstrated an impaired acute insulin response and insulin action in obese Caucasian women with former GDM, while Ryan *et al.*[8] showed defective insulin action in non-obese Caucasian women with former GDM and normal glucose tolerance. We do not have a concrete evidence to explain these discrepancies. However, Bergstrom *et al.*[28] and Kahn *et al.*[29] have found impaired β-cell function in second generation Japanese–Americans before the development of Type 2 DM. Taken together, these studies indicate that the pathogenesis of Type 2 DM is influenced by environmental factors and ethnic and genetic factors.

We used 20 mU kg^{-1} of insulin during FSIGT, less than the dosage of 30 mU kg^{-1} used in other studies. Although the level of insulin attained is important for

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the minimal model to function appropriately, the smaller dosage of insulin probably is unlikely to explain our current findings. The 20 mU kg^{-1} of insulin has been employed before by us and by others.[17,25,26,30]

In summary, we have demonstrated that in non-obese Japanese women with former GDM, glucose tolerance (Kg), acute insulin secretion, and glucose effectiveness at zero insulin were significantly lower than normal, although insulin sensitivity was normal. These results indicate that deterioration of acute β-cell responses to glucose and tissue glucose sensitivity rather than tissue insulin sensitivity may be pathogenic factors in the development Type 2 DM in Japanese former GDM.

Acknowledgements

We would like to acknowledge K. Tokuyama (Laboratory of Biochemistry of Exercise and Nutrition, Institute of Health and Sports Sciences, University of Tsukuba) for the technical instruction for minimal model analysis.

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